

细胞内钾离子稳态测量技术及其调控机制研究进展

颜芷昕^{1,2} 宋娜娜^{1,2,3} 丁小强^{1,2△}

(¹复旦大学附属中山医院肾内科 上海 200032; ²上海市肾病与血液透析重点实验室 上海 200032;

³复旦大学张江研究院 上海 200120)

【摘要】 钾离子在细胞内液、细胞外液及亚细胞结构中的分布处于动态平衡,它是细胞内液含量最丰富的阳离子,在细胞外液含量却很少;在亚细胞结构中,钾离子在胞核的浓度最高,细胞基质、线粒体、内质网、溶酶体次之。细胞内钾离子参与细胞形态维持、酶活性调控及能量代谢、遗传物质的生物合成、细胞增殖与死亡、修复和迁移、免疫等多种生命活动的调控;同时细胞内钾离子稳态失衡和肿瘤、急性缺血损伤、自身免疫病、内分泌、神经退行性等疾病发生发展密切相关,因此维持细胞内钾离子稳态具有非常重要的意义,纠正细胞内钾离子稳态失衡为疾病治疗提供了新的方向。本文综述近年细胞内钾离子稳态的基本情况和测量技术的研究进展,并探讨其调控机制及生理和病理意义。

【关键词】 细胞内钾离子稳态; 细胞内钾离子浓度检测; 钾离子敏感荧光探针; 基因编码钾离子指示剂; 钾离子通道

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Research progress of measuring techniques and regulation mechanisms of intracellular potassium homeostasis

YAN Zhi-xin^{1,2}, SONG Na-na^{1,2,3}, DING Xiao-qiang^{1,2△}

(¹Department of Nephrology, Zhongshan Hospital, Fudan University, Shanghai 200032, China; ²Shanghai Key Laboratory of Kidney and Blood Purification, Shanghai 200032, China; ³Fudan ZhangJiang Institute, Shanghai 200120, China)

【Abstract】 Distribution of potassium ion (K^+) among extracellular/intracellular fluid and subcellular organelles is in dynamic equilibrium under exquisite manipulation. K^+ is most abundant in nuclei, which is followed by cytoplasmic matrix, mitochondria, endoplasmic reticulum and lysosome. Intracellular potassium homeostasis has great significance. It plays important roles in cell morphology maintenance, enzyme activities and energy metabolism modulation, biosynthesis of genetic material, cell proliferation and death, cell repair and migration, and immune response. Also, intracellular potassium disturbance is closely related to tumor, acute ischemic injuries, autoimmune diseases, endocrine diseases and neurodegenerative diseases. Correcting intracellular potassium disturbance has become a novel direction in clinical therapy. We give an overview of the current physiological condition, measuring techniques, regulation mechanisms and physiological/pathological significances of intracellular potassium homeostasis.

【Key words】 intracellular potassium homeostasis; intracellular potassium concentration measurement; potassium-sensitive fluorescent probes; genetically encoded potassium indicators;

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[△]Corresponding author E-mail: ding.xiaoqiang@zs-hospital.sh.cn

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potassium channel

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维持机体钾平衡是保证多种生命活动正常运行的必要条件,钾平衡紊乱会导致多系统、多器官的异常,机体钾缺乏可促心血管疾病^[1]、慢性肾脏病^[2]的发生和发展。临床上一般用血钾和尿钾水平来评估机体钾的平衡状态,这些指标主要反映细胞外液钾离子水平。然而,越来越多的研究提示细胞外液钾离子浓度不够灵敏和真实地反应机体钾水平,如长期低钾饮食可使血钾浓度依旧接近正常值,但此时机体的低钾状态已造成肾损伤指标的升高^[3],因此需要一个更真实更灵敏的指标反映机体钾水平——细胞内钾离子浓度可能更为合适。钾离子在细胞内外并非呈均衡分布,细胞内液钾离子浓度(150 mmol/L)约是细胞外液(3.5~5 mmol/L)的30倍^[4]。在慢性肾脏病的模型中,尽管此时的血钾浓度可能并无明显改变,但细胞内钾离子浓度明显减少,及时纠正细胞内低钾的情况可改善细胞周期阻滞及肾脏纤维化^[5],细胞内液钾离子可能作为细胞外液的缓冲系统,较细胞外液先发生平衡紊乱,因此细胞内液钾离子水平更灵敏地反映机体钾状态。此外,维持细胞内钾离子稳态可调节能量代谢、细胞增殖和死亡等过程^[6-7],维持细胞内钾离子稳态具有重要的意义。

细胞内钾离子稳态基本情况 细胞内钾离子稳态有两个层面:一是细胞内液总钾平衡,二是亚细胞结构钾离子的分布平衡。囿于技术限制,关于细胞器中钾离子的分布以及细胞内钾离子浓度在不同生理和病理条件下的动态变化一直未有定论,但随着近几年关于靶向亚细胞结构的荧光探针、基因编码钾离子指示剂及光学技术的发展,研究者在几个不同类别的细胞中均发现:胞核的钾离子浓度最高(150~350 mmol/L)且细胞核基质的浓度高于核周间隙,细胞基质的钾离子浓度居中(50~100 mmol/L),线粒体(40~50 mmol/L)、内质网(2~8 mmol/L)和溶酶体(2~50 mmol/L)的钾离子浓度较低^[8-10],需要注意的是不同类型的细胞、不同病理状态下细胞内钾离子情况并不相同。

细胞内钾离子浓度的测定 长久以来,对细胞内钾离子浓度的测定是研究细胞内钾离子相关生

理病理研究的瓶颈,直接测量难度较大、操作繁琐,更多的研究是采用间接测量(电生理)的方法,通过对离子通道电流的分析间接推断细胞内钾离子的情况(表1)。

原子吸收光谱法 将细胞裂解后对裂解液进行光谱分析,准确度高且可定量,但具有细胞破坏性。

核磁共振波谱法(³⁹K-nuclear magnetic resonance, ³⁹K-NMR) 据核磁共振的原理测量钾,操作时收集待测细胞至特殊小管,根据得到的谱图进行分析^[11],此方法无细胞损伤性,但灵敏度和精确度低且难以获得亚细胞结构的分布信息。

钾离子选择性玻璃电极法 玻璃电极由钾离子选择电极和参比电极共同组成,其中钾离子选择电极含特殊的含缬氨霉素的聚氯乙烯膜成分,当电极破坏细胞膜和细胞内液接触后,可在聚氯乙烯膜上产生电位并与参比电极构成回路获得电极电位,以此计算出细胞内钾离子浓度。此方法准确、敏感度高^[12],但具有细胞损伤性且无法获取钾离子在细胞内的时空分布情况。

合成钾离子敏感的荧光染料法 荧光探针可选择性螯合细胞内游离钾离子,当钾离子与探针结合后可改变探针的构象,增加其量子产率,使光谱发生偏移,通过分析产生的荧光信号可获得细胞内钾离子浓度的数据。理想的钾离子荧光探针应满足对生理条件下细胞内钾离子浓度变化的敏感性以及选择特异性。PBFI是应用最早和最广为人知的钾离子荧光探针之一,但是它对钾离子的选择性差,易受到钠离子干扰,而且操作繁琐。后来研究者研制出钾离子选择性更高的探针,如KS2探针^[13]和可降低假阳性率并用于活细胞成像的双荧光探针^[14-15];还有能定位于亚细胞结构的钾离子探针,如半定量检测线粒体内钾离子浓度变化的KS6探针^[16]。

基因编码的钾离子指示剂 研究者发现生物体内存在可螯合钾离子的钾离子选择性蛋白,为了改善化学合成荧光探针有一定细胞毒性、特异性差、低通量的缺点,他们提出可采用基因工程的方法,通过在原核生物体内共表达和提纯具有高度钾

离子选择性蛋白和荧光蛋白进而获得基因编码的钾离子指示剂,并认为此方法更安全、高效^[8]。此外,在钾离子指示剂上添加细胞器特异序列可实现靶向亚细胞的钾离子动态监测,推进对细胞器钾离子功能的研究,目前有靶向线粒体的KS6^[16]、Mt lc-LysM GEPII 钾离子探针^[8]以及靶向细胞核的

NUC1c-LysMGEPII 钾离子探针^[8],这些探针具有高灵敏性、特异性、时空分布性、高通量的优势。目前基于双荧光能量共振转移发光方式的指示剂有:GEPII 系列^[8]、KIRIN1-GR^[17];基于单荧光的有GINKO,和双荧光发光方式相比,单荧光团体积小、适用于多荧光检测^[17](表1)。

表1 细胞内钾离子浓度测量方法
Tab 1 Measuring methods of intracellular K⁺ concentration

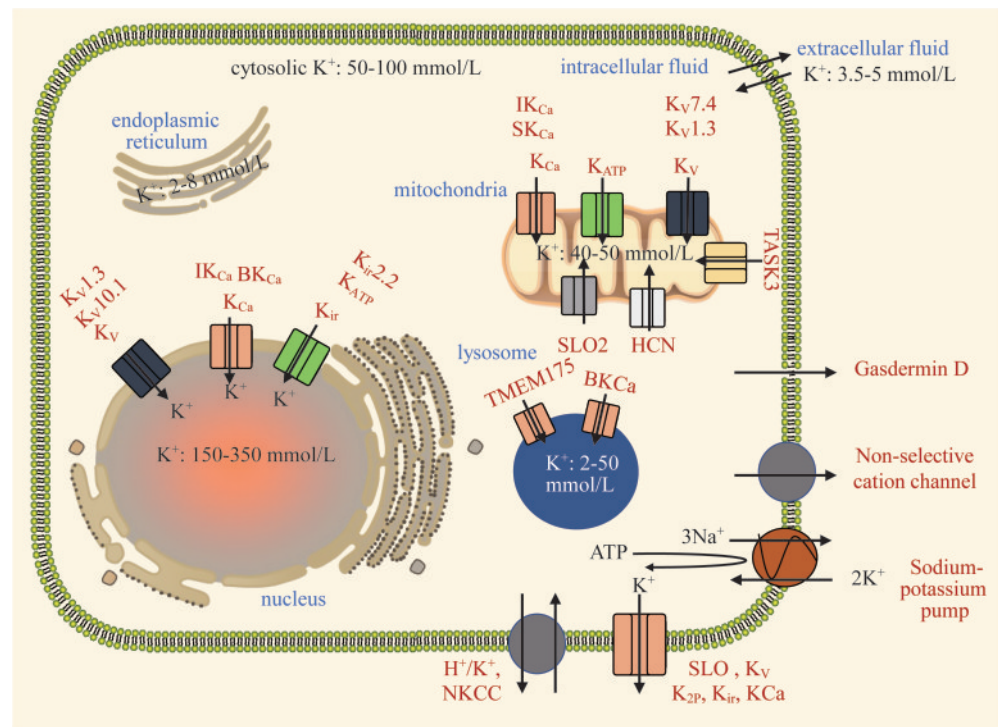
Category		Advantages	Disadvantages
Apply to cytosolic K ⁺ concentration measurement			
Atomic absorption spectrophotometry	—	Highly accurate and specific; Quantitative	Invasive; Unable for real time measurement
³⁹ K-NMR ^[11, 18-19]	—	Non-invasive	Poorly accurate; Unable for real time measurement
Potassium ion specific electrode ^[12]	—	Highly accurate	Invasive; Unable for real time measurement
Selective fluorescent potassium sensor	PBFI ^[20]	Easily accessible	Non-specific
	KS2 ^[13] , NIR ^[14] , FRET ^[15]	Highly specific; Suitable for real time and live cell measurement	Tedious
Genetically encoded fluorescent potassium indicator	GEPII 1.0 ^[8] , GINKO KIRIN1 ^[17]	Highly specific; Suitable for real time and live cell measurement	Hardly accessible
Apply to mitochondrial K ⁺ concentration measurement			
Selective fluorescent potassium sensor	KS6 ^[16] , TAC-Rh ^[21]	Highly selective labeling of mitochondria	—
Genetically encoded fluorescent potassium indicator	Mt lc-LysM GEPII 1.0 ^[8]		—
Apply to nuclear K ⁺ concentration measurement			
Genetically encoded fluorescent potassium indicator	NUC1c-LysMGEPII1.0 ^[8]	Highly selective labeling of nucleus	—

细胞内钾离子稳态的调控机制 由于钾离子不能自由进出细胞膜结构,细胞内液钾离子浓度稳态依赖于细胞膜上特殊通道的开放程度和细胞内外钾离子电化学梯度;而亚细胞结构的钾离子分布平衡则依赖于细胞器膜上的特殊通道(图1)。

细胞内总钾平衡 钾离子在细胞内外形成的电化学梯度和膜上的孔道性质共同决定钾离子的进出方向,细胞膜上钾离子流动的孔道主要有:钾特异性通道、钾离子交换体、钠钾泵、非选择性阳离子通道和特殊“膜孔”结构。(1)钾通道是钾离子流动最常见的孔道,根据门控机制、结构和功能主要分成了5个家族:电压门控钾通道K_v;内向整流钾通道K_{ir};钙和钠激活大电导钾通道SLO;小电导钙激活钾通道SKCa以及双孔钾通道K_{2p}^[22]。由于胞内钾离子浓度明显高于胞外,大多钾通道在一般生理条件下顺电化学梯度介导钾离子由胞内流向胞外。(2)钾离子交换体和转运体:如氢-钾交换体(钾离子进出方向取决于两种离子在细胞内外的浓度

差,酸中毒时氢离子转运入胞内,钾离子转运出胞外)和钠钾二氯同向转运体(介导钾离子内流入胞)等;(3)钠钾泵:将钾离子逆浓度梯度转运至胞内,维持胞内高钾环境;(4)非选择性阳离子通道:如瞬时受体电位阳离子通道亚家族(transient receptor potential channel, TRPC),介导钾离子由胞内流向胞外;(5)特殊膜孔:焦亡效应蛋白gasdermin D在胞膜上形成孔结构,结合钾离子并介导钾离子外流^[23]。当钾离子由胞内流向胞外时,细胞内钾离子浓度减小;反之,细胞内钾离子浓度增多。改变细胞外液钾离子浓度,如增加细胞外钾离子浓度可减弱电化学梯度,减小钾离子外流,使胞内钾浓度增多。

亚细胞结构间钾离子分布的平衡 钾离子在细胞内并非均匀分布,在钾通道的调节下维持亚细胞结构间钾离子分布平衡(表2),胞核钾离子浓度最高,细胞基质次之,其他细胞器(线粒体、溶酶体、内质网等)浓度最低。目前亚细胞结构的膜上的钾离子通道研究仍非常有限,一般认为钾离子的流动



Intracellular potassium homeostasis is regulated by the dynamic equilibrium of influx, efflux and the intracellularly stored amount of K^+ . K^+ is most abundant in nuclei, which is followed by cytoplasm, mitochondria, lysosome and endoplasmic reticulum. Potassium channels/potassium ion exchangers/sodium-potassium pump/non-selective cation channels in plasma membrane and specific plasma membrane pores (gasdermin D, e.g.) contribute to potassium ions flux between extracellular fluid and intracellular fluid, while potassium channels located at membranes of intracellular organelles play roles in potassium ions influx and subcellular potassium distribution.

图1 细胞内钾离子稳态的基本情况与调控机制

Fig 1 Intracellular K^+ distribution and machinery for K^+ regulation

方向是由胞浆流入线粒体、内质网、溶酶体和细胞核基质。细胞核维持高钾离子水平的机制,细胞器膜上是否存在其他钾离子孔道(如交换体等)待进一步研究。

表2 亚细胞结构中钾离子分布调控

Tab 2 Regulation mechanisms about distribution of K^+ among subcellular structure

Family (channel name)		Location	Mechanism
K_{ir}	K_{ATP}	The inner mitochondrial membrane	With channels opening, K^+ flows from the cytosol to mitochondria which increases mitochondrial K^+ concentration
K_V	$K_V1.3; K_V7.4$		
K_{Ca}	$IK_{Ca}; SK_{Ca}$		
K_{2p}	TASK3		
SLO	SLO2;		
HCN	—		
—	TMEM175	Lysosome membrane	With channels opening, K^+ flows from the cytosol to lysosome which increases lysosomal K^+ concentration
K_{Ca}	BKCca		
K_{Ca}	BKCca; IK_{Ca}	Nuclear envelope	With channels opening, K^+ flows from the cytosol to nucleus which increases nuclear K^+ concentration
K_{ir}	K_{ATP}		
K_V	$K_V1.3; K_V10.1$		
K_{ir}	$K_{ir}2.2$		

Potassium channels in the membranes of nucleus/mitochondria/lysosome contribute to influx of K^+ into these organelles.

钾离子通道的调控机制 由于钾通道对钾离子的高度特异性,钾通道在细胞内钾离子稳态调控占主体地位。目前已发现的钾通道有80多种,每种钾通道的调控机制各不相同。总体而言,主要包括通道本身的门控机制(电压、机械张力、ATP、pH等)和信号分子对通道活性和表达水平的调控(如炎症、氧化应激相关信号分子)。

门控机制 作为钾离子通道本身的特性,钾离子通道的开放可由膜电位/电压(如电压门控钾通道 K_v 家族)、胞内外pH(如酸敏感的双孔钾通道TASK2)、机械刺激(如弱内向整流相关双孔钾通道TREK1)、ATP水平(如ATP敏感的钾通道 K_{ATP})、胞内其他阳离子(Ca^{2+} 、 Mg^{2+} 、 Na^+)浓度、G蛋白偶联受体激活的G蛋白信号通路(如被激素激活)等调控。

信号通路 (1)炎症相关信号通路:炎症过程中产生的细胞因子可调控钾通道活性, γ 干扰素(interferon- γ , IFN- γ)通过与细胞膜上的受体结合,在近端肾小管细胞内产生过氧化物或过氧化亚硝基,使钾离子通道发生氧化或亚硝基化进而被抑制;白细胞介素 1β (interleukin 1β , IL- 1β)可激活蛋白激酶C(protein kinase C, PKC)通路,抑制钾离子通道^[24]。(2)氧化代谢相关信号通路:多种钾通道和氧化应激或氧化代谢通路相关。缺氧诱导因子(hypoxia-inducible factor, HIF)是氧感受通路的关键转录因子,研究发现缺氧通过增加HIF-1上调钾通道表达水平,如在肺动脉平滑肌细胞中上调 BK_{Ca} 表达^[25]以及在B淋巴细胞中上调TASK2表达^[26]。活性氧(reactive oxygen species, ROS)是缺氧过程中线粒体代谢产物,线粒体ATP敏感的钾通道常在缺氧的条件下被ROS产物活化^[27]。ROS还可氧化修饰钾通道进而改变通道活性, BK 通道位于细胞内的3个蛋氨酸被ROS氧化修饰后活性增强而半胱氨酸被氧化修饰后活性减弱^[28]。细胞内钙离子浓度增加也常见于氧化应激过程,可激活钙敏感钾通道的开放。氧化应激过程还可通过激活cAMP/GMP依赖的蛋白激酶信号通路磷酸化钾通道亚基,进而改变通道的活性。

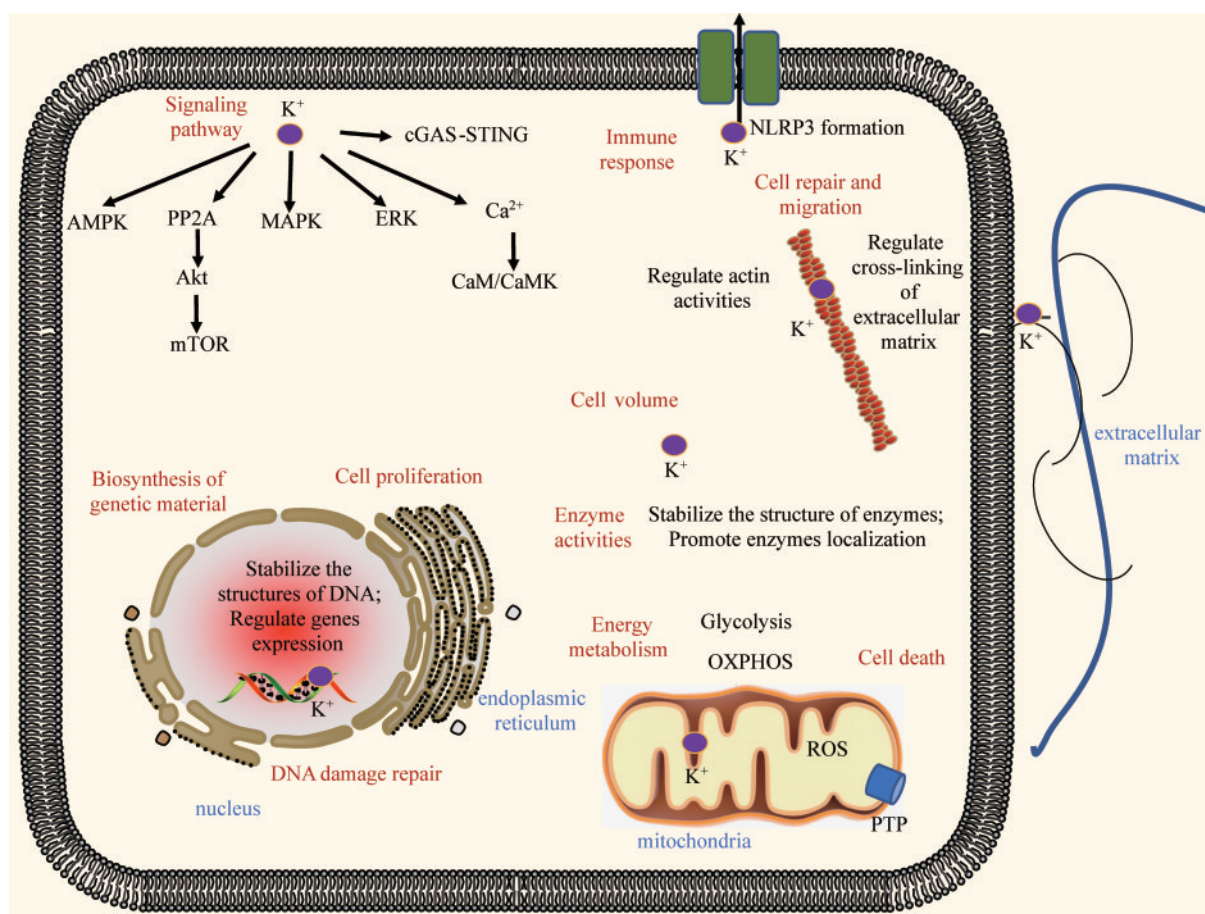
细胞内钾离子稳态的生理作用 细胞内钾离子基本功能是维持细胞静息膜电位,这是可兴奋细胞正常发挥其兴奋功能的必要条件。此外,钾离子还参与调控多种生命活动(图2)。

调节酶及能量代谢 细胞内钾离子通过稳定酶结构维持酶活性^[29],如丙酮酸激酶、丙酮酸脱氢酶、DNA双链断裂修复蛋白RAD51等;也可作为酶的辅因子,如钾离子是己糖激酶II的辅因子,促葡萄糖分解^[6]。此外,细胞内钾稳态参与细胞能量代谢过程:胞内钾离子浓度降低时,AMP依赖的蛋白激酶通路活化,糖酵解过程被抑制,线粒体ATP生成减少,线粒体氧化磷酸化功能受抑制^[6]。激活线粒体钾通道,促线粒体内膜去极化,促呼吸链运行和ATP生成,脂肪酸氧化过程加强^[30]。

调节遗传物质的生物合成 细胞内钾离子可与DNA结合,稳定核酸G-四链体结构,调节DNA复制^[31];胞内钾离子参与调节转录因子的转录活性;抑制核膜上的钾离子通道开放(如 BK_{Ca} 、 $K_v1.3$ 等),促核膜超极化,最终导致环磷腺苷效应元件结合蛋白(cAMP response element-binding protein, CREB)磷酸化,促进c-fos等极早期反应蛋白转录^[10,32]。

调节细胞容积和形态 细胞内钾离子维持细胞容积的稳态:钾离子是胞内浓度最高的阳离子,是维持细胞内渗透压的主要组成成分;而细胞内外的渗透压差决定细胞液体流动方向,影响细胞的容积和形态。凋亡相关细胞容积减小与钾离子通道激活介导的钾离子外流相关^[33];当细胞过度肿胀时钾离子通道也会被激活,进而降低细胞内钾离子浓度,减小细胞内的渗透压,抑制肿胀进一步加剧^[34]。

调节细胞周期和增殖 增殖是细胞的基本功能,其中细胞周期的精准调控保障增殖的有序进行。最初研究者发现非兴奋性细胞的细胞周期中伴随着膜电位的周期性变化,G1期→S期超极化,G2期→M期去极化^[35-36];而钾离子是维持静息膜电位的主要离子,研究进一步发现细胞内钾离子浓度的改变可直接影响细胞周期和增殖,抑制外周血淋巴细胞钾离子通道活性或增加细胞外液钾离子浓度进而提高细胞内钾离子浓度,可抑制淋巴细胞增殖和分泌细胞因子^[36];我们团队发现体外诱导纤维化的近端肾小管上皮细胞以及体内纤维化肾脏模型中细胞内钾离子浓度减低,并促进上皮细胞G2/M期阻滞^[5]。同时,钾电流和钾离子通道的表达水平可随细胞周期变化^[37],钾通道是调控细胞内钾离子稳态的重要手段,钾离子通道的开放通过影响细胞内钾稳态进而调节细胞周期。



Intracellular potassium homeostasis plays diverse roles in biological processes including cell morphology maintenance, enzyme activities and energy metabolism modulation, biosynthesis of genetic material, cell proliferation and death, cell repair and migration, and immune response. OXPHOS: Oxidative phosphorylation; PTP: Permeability transition pore.

图2 细胞内钾离子稳态的生理功能

Fig 2 Physiological roles of intracellular potassium homeostasis

然而细胞内钾离子调控细胞周期的具体机制尚缺乏统一的认识,目前有如下几种学说:(1)钙学说:细胞内钾离子浓度的减低为钙离子内流提供驱动力,促使细胞内钙离子浓度的升高,钙离子作为第二信使启动 Ca^{2+} —钙调蛋白/钙调素依赖蛋白激酶 (calmodulin/calmodulin kinase, CaM/CaMK) 通路调控细胞周期关键蛋白,比如上调细胞周期抑制因子 p27、p21 等表达,阻滞细胞周期^[37]; (2)信号通路学说:钾离子通过调控 cAMP 依赖蛋白激酶 A (protein kinase A, PKA)、P38 丝裂原活化蛋白激酶 (P38 mitogen activated protein kinases, P38-MAPK)、细胞外调节蛋白激酶 (extracellular signal regulated kinase, ERK) 以及蛋白激酶 B (Akt) 相关信号通路改变细胞周期蛋白的表达水平^[38],但需更多的研究证实。

调节细胞死亡 胞内钾稳态调控与凋亡的发

生密切相关,凋亡的其中一个特征是细胞容积减小,而钾离子作为胞内含量最高的离子对细胞容积的调控至关重要,胞膜钾通道的激活、钾离子外流和细胞内钾离子浓度降低均可促进凋亡发生^[33]。线粒体内膜上的钾通道和线粒体钾离子稳态也参与调控死亡,抑制线粒体钾离子通道 (TASK3、 K_v 等) 可促进 ROS 释放、抑制呼吸链并诱导死亡^[30]; 还有研究提示激活线粒体钾离子通道 SK、增加线粒体钾浓度可减轻细胞铁死亡^[39]。

调节细胞迁移和修复 细胞 (尤其是表皮细胞) 迁移是发生损伤修复的重要手段之一,迁移过程包括细胞骨架重构、细胞膜突出、前部的细胞外基质的局部黏附和后部的细胞外基质去黏附。研究者发现细胞迁移过程中细胞膜上的钾离子通道分布不均匀,如肾上皮细胞发生迁移时, $\text{K}_v1.4$ 通道聚集在板状伪足的前部突出部分,抑制这部分的钾

离子通道会抑制细胞迁移,而 K_{Ca} 通道分布在胞体尾部,抑制这部分的离子通道同样会抑制细胞迁移^[40];迁移相关信号通路如受体蛋白酪氨酸激酶(如表皮生长因子及受体)及非受体蛋白激酶(如ERK)信号通路的激活可上调钾离子通道的表达、增加钾离子外流进而促进细胞迁移。同时迁移信号通路也受钾离子通道调控,抑制钾离子通道可抑制迁移信号通路^[41]。

目前钾离子调控细胞迁移修复的机制包括:(1)钾通道开放、钾离子的外流为钙离子内流提供驱动力,通过激活钙信号通路促进细胞迁移^[42];(2)细胞内钾离子直接可作为第二信使调控迁移相关蛋白合成^[43],也可调节局部渗透压与容积,调控肌动蛋白促进细胞骨架重构^[44];(3)通过改变局部离子浓度影响细胞外基质组装、交联过程^[42,45]。

调节免疫应答 细胞内钾离子通过炎症小体(NLR family pyrin domain containing 3, NLRP3)活化、炎症因子生成和免疫细胞功能调节参与机体的免疫应答。NLRP3介导的炎症反应常伴随着细胞内钾离子外流、细胞内钾离子浓度减低的现象^[46],细胞内钾离子外流促进NLRP3进一步形成,激活细胞死亡的过程^[23,47]。细胞内钾离子外流可抑制DNA感受器环GMP-AMP合酶-干扰素刺激基因(cyclic GMP-AMP and the stimulator of interferon genes, cGAS-STING)反馈通路,调控I型干扰素的生成^[23]。免疫细胞(T/B/NK细胞)上的钾离子通道开放及其介导的细胞内钾离子外流还可通过膜超极化为钙离子的内流提供驱动力、增加细胞内钙离子浓度,钙离子作为第二信使可活化转录因子进而诱导下游基因转录促免疫细胞成熟、分化及发挥效应^[48-49],如钾通道 $K_V1.3$ 和 $IK_{Ca}2$ 的激活可促活化T细胞核因子(nuclear factor of activated T cells, NFAT)的核定位并启动下游基因转录,促T细胞增殖和分泌细胞因子^[50]。

细胞内钾离子稳态失衡和疾病的联系

肿瘤 在多种肿瘤(如乳腺癌、宫颈癌、神经胚胎瘤、结肠癌、急性髓性白血病等)存在钾通道表达异常^[51-53],研究进一步发现肿瘤的组织间液的钾离子浓度升高^[7],二者均可影响细胞内钾离子稳态,细胞内钾失衡和肿瘤密切相关。细胞内钾稳态失衡促肿瘤发生发展的机制复杂多样,涉及细胞增殖死亡、损伤修复和免疫功能异常。细胞内钾稳态失衡

破坏细胞周期的有序进行,细胞周期蛋白(如cdc25、cdc2)表达异常^[54],促肿瘤细胞不断分裂增殖;钾离子通道(如 K_{Ca} 家族、 K_V 通道家族)活化可促肿瘤细胞DNA损伤修复,增强肿瘤细胞对放化疗的抵抗性^[55],高表达钾离子通道的肿瘤组织可能提示不良预后;细胞内钾离子还参与调节肿瘤免疫,肿瘤微环境的钾离子浓度高于正常组织的细胞外液,造成其中的T细胞内钾离子浓度升高,增强磷酸酶PP2A的活性并抑制蛋白激酶B-雷帕霉素靶蛋白(Akt-mTOR)通路磷酸化程度,最终抑制T细胞发挥效应,削弱对肿瘤细胞的杀伤作用^[7]。

急性缺血相关疾病 细胞膜钾通道(K_V 、 K_{ATP} 、TREK1等)开放可通过抑制钙超载、促DNA损伤修复和减轻凋亡在多种急性缺血性脏器损伤中发挥保护作用,包括急性心肌梗死^[56]、卒中^[57]、急性缺血再灌注肾小管损伤^[58]等。线粒体内膜钾通道(如BK)的开放可减轻缺血诱导的心肌损伤——阻断或抑制线粒体BK通道可抑制线粒体呼吸链,增加ROS产生,增加细胞凋亡,最终加剧心肌损伤^[59-60]。

自身免疫疾病 钾通道的异常表达(增多或减低)、异常功能及其介导的细胞内钾离子稳态失衡和自身免疫疾病的发生发展密切相关,包括系统性红斑狼疮^[61]、多发性硬化^[62]、炎症性肠病^[62-63]、类风湿性关节炎^[64]等,其中的病理生理机制尚未明确。自身免疫疾病模型中的免疫细胞(B/T/NK细胞)常见钾离子通道表达上调、功能过度活化以及细胞内钾离子浓度减低,细胞内钾离子浓度异常减低可促进免疫细胞的异常活化,而免疫细胞的过度激活是自身免疫疾病的重要发病机制之一^[65];而在病变脏器常见钾离子通道表达减少、通道活性受抑制,最终抑制组织器官发挥功能^[66]。

内分泌疾病 内分泌器官的钾离子通道和细胞内钾离子稳态维持内分泌激素的正常分泌,钾离子通道表达减少、功能缺失可见于糖尿病(新生儿糖尿病^[67]、2型糖尿病^[68])以及高醛固酮血症^[69],并造成激素分泌紊乱。肾上腺钾通道(如TASK1、TASK3)的缺失造成细胞内钾离子浓度升高、细胞持续去极化,细胞兴奋周期被破坏、钙离子内流增强,不断合成和分泌醛固酮激素^[70]。

神经退行性疾病 近年研究者发现钾通道表达减少和功能受损常见于神经退行性疾病,包括亨廷顿病^[71]、帕金森病^[72]、阿尔兹海默病^[73]等,并可作

为疾病治疗的重要靶点之一。胞膜上的钾通道表达减少、细胞内钾离子浓度升高可直接导致神经系统的兴奋毒性、细胞受损和神经环路的破坏^[74];激活线粒体钾通道如K_{ATP}可减轻线粒体损伤、保护胆碱能中间神经元,最终缓解亨廷顿病^[75];TMEM175是钾离子进出溶酶体的主要通道并维持溶酶体功能^[76],TMEM175表达减少与功能缺失可通过抑制溶酶体功能,加剧 α -突触核蛋白的异常沉积、多巴胺神经元的减少和运动功能损害,最终促进帕金森病的进展^[77]。

结语 钾离子主要分布于细胞内液,细胞内钾离子稳态的改变往往先于细胞外液。细胞内钾稳态由细胞内、外液和细胞器间钾离子分布平衡共同维持,参与细胞生长死亡、黏附迁移、能量代谢和免疫调控等多种活动。细胞内钾离子稳态失衡与肿瘤、急性缺血、自身免疫、内分泌、神经退行性疾病的发生发展密切相关,并提示疾病进展、预后,靶向干预钾离子通道以及纠正细胞内电解质水平为临床提供了新的治疗思路。受测量细胞内钾离子浓度检测技术的限制,先前大多数研究都是通过钾通道的功能变化间接推测细胞内钾离子浓度的改变,缺乏细胞内钾稳态失衡与疾病发生发展相关性的直接证据,总体上对胞内钾离子的调控机制及在细胞功能调控中的作用机制的认知是远远不够的。近年钾离子敏感的荧光探针和基因编码的钾离子指示剂技术手段的发展将进一步促进人们对细胞内钾离子稳态调控机制的探索,并为疾病的诊疗提供新的方向。

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参 考 文 献

- [1] MCDONOUGH AA, VEIRAS LC, GUEVARA CA, et al. Cardiovascular benefits associated with higher dietary K⁺ vs. lower dietary Na⁺: evidence from population and mechanistic studies [J]. *Am J Physiol Endocrinol Metab*, 2017, 312(4): E348-E356.
- [2] PICARD K, BARRETO SILVA MI, MAGER D, et al. Dietary potassium intake and risk of chronic kidney disease progression in predialysis patients with chronic kidney disease: a systematic review [J]. *Adv Nutr*, 2020, 11(4): 1002-1015.
- [3] TERKER AS, ZHANG Y, ARROYO JP, et al. Kir4.2 mediates proximal potassium effects on glutaminase activity and kidney injury [J]. *Cell Rep*, 2022, 41(12): 111840.
- [4] PALMER BF. Regulation of potassium homeostasis [J]. *Clin J Am Soc Nephrol*, 2015, 10(6): 1050-1060.
- [5] ZHANG J, CHEN J, LU Y, et al. TWIK-related acid-sensitive K⁺ channel 2 promotes renal fibrosis by inducing cell-cycle arrest [J]. *iScience*, 2022, 25(12): 105620.
- [6] BISCHOF H, BURGSTALLER S, SPRINGER A, et al. Potassium ions promote hexokinase- II dependent glycolysis [J]. *iScience*, 2021, 24(4): 102346.
- [7] EIL R, VODNALA SK, CLEVER D, et al. Ionic immune suppression within the tumour microenvironment limits T cell effector function [J]. *Nature*, 2016, 537(7621): 539-543.
- [8] BISCHOF H, REHBERG M, STRYECK S, et al. Novel genetically encoded fluorescent probes enable real-time detection of potassium *in vitro* and *in vivo* [J]. *Nat Commun*, 2017, 8(1): 1422.
- [9] BISCHOF H, BURGSTALLER S, WALDECK-WEIERMAIR M, et al. Live-cell imaging of physiologically relevant metal ions using genetically encoded fret-based probes [J]. *Cells*, 2019, 8(5): 492.
- [10] BURGSTALLER S, BISCHOF H, MATT L, et al. Assessing K⁺ ions and K⁺ channel functions in cancer cell metabolism using fluorescent biosensors [J]. *Free Radic Biol Med*, 2022, 181: 43-51.
- [11] ADAM WR, KORETSKY AP, WEINER MW. Potassium adaptation: 39K-NMR evidence for intracellular compartmentalization of K⁺ [J]. *Am J Physiol*, 1988, 254(3Pt 2): F401-F406.
- [12] FRANT MS, ROSS JW, JR. Potassium ion specific electrode with high selectivity for potassium over sodium [J]. *Science*, 1970, 167(3920): 987-988.
- [13] ZHOU X, SU F, TIAN Y, et al. A new highly selective fluorescent K⁺ sensor [J]. *J Am Chem Soc*, 2011, 133(46): 18530-18533.
- [14] SUI B, YUE X, KIM B, et al. Near-IR two-photon fluorescent sensor for K⁺ imaging in live cells [J]. *ACS Appl Mater Interfaces*, 2015, 7(32): 17565-17568.
- [15] YANG Y, HUANG J, YANG X, et al. Aptamer-based FRET nanoflares for imaging potassium ions in living cells [J]. *Chem Commun (Camb)*, 2016, 52(76): 11386-11389.
- [16] KONG X, SU F, ZHANG L, et al. A highly selective

- mitochondria-targeting fluorescent K^+ sensor [J]. *Angew Chem Int Ed Engl*, 2015, 54(41):12053-12057.
- [17] SHEN Y, WU SY, RANCIC V, *et al.* Genetically encoded fluorescent indicators for imaging intracellular potassium ion concentration [J]. *Commun Biol*, 2019, 2:18.
- [18] BROPHY PJ, HAYER MK, RIDDELL FG. Measurement of intracellular potassium ion concentrations by n.m.r [J]. *Biochem J*, 1983, 210(3):961-963.
- [19] ADAM WR, KORETSKY AP, WEINER MW. Measurement of tissue potassium *in vivo* using ^{39}K nuclear magnetic resonance [J]. *Biophys J*, 1987, 51(2):265-271.
- [20] MEUWIS K, BOENS N, DE SCHRYVER FC, *et al.* Photophysics of the fluorescent K^+ indicator PBFI [J]. *Biophys J*, 1995, 68(6):2469-2473.
- [21] SONG G, JIANG D, WANG L, *et al.* A mitochondria-targeting NIR fluorescent potassium ion sensor: real-time investigation of the mitochondrial K^+ regulation of apoptosis *in situ* [J]. *Chem Commun (Camb)*, 2020, 56(40):5405-5408.
- [22] GONZALEZ C, BAEZ-NIETO D, VALENCIA I, *et al.* K^+ channels: function-structural overview [J]. *Compr Physiol*, 2012, 2(3):2087-2149.
- [23] BANERJEE I, BEHL B, MENDONCA M, *et al.* Gasdermin D restrains type I interferon response to cytosolic DNA by disrupting ionic homeostasis [J]. *Immunity*, 2018, 49(3):413-426.e5.
- [24] NAKAMURA K, HAYASHI H, KUBOKAWA M. Proinflammatory cytokines and potassium channels in the kidney [J]. *Mediators Inflamm*, 2015, 2015:362768.
- [25] AHN YT, KIM YM, ADAMS E, *et al.* Hypoxia-inducible factor-1 α regulates KCNMB1 expression in human pulmonary artery smooth muscle cells [J]. *Am J Physiol Lung Cell Mol Physiol*, 2012, 302(3):L352-L359.
- [26] SHIN DH, LIN H, ZHENG H, *et al.* HIF-1 α -mediated upregulation of TASK-2 K^+ channels augments Ca^{2+} signaling in mouse B cells under hypoxia [J]. *J Immunol*, 2014, 193(10):4924-4933.
- [27] QUELICONI BB, WOJTOVICH AP, NADTOCHIY SM, *et al.* Redox regulation of the mitochondrial K(ATP) channel in cardioprotection [J]. *Biochim Biophys Acta*, 2011, 1813(7):1309-1315.
- [28] SESTI F, LIU S, CAI SQ. Oxidation of potassium channels by ROS: a general mechanism of aging and neurodegeneration? [J]. *Trends Cell Biol*, 2010, 20(1):45-51.
- [29] GOHARA DW, DI CERA E. Molecular mechanisms of enzyme activation by monovalent cations [J]. *J Biol Chem*, 2016, 291(40):20840-20848.
- [30] CHECCHETTO V, LEANZA L, DE STEFANI D, *et al.* Mitochondrial K^+ channels and their implications for disease mechanisms [J]. *Pharmacol Ther*, 2021, 227:107874.
- [31] HOWARD JJ, LYNCH GC, PETTITT BM. Ion and solvent density distributions around canonical B-DNA from integral equations [J]. *J Phys Chem B*, 2011, 115(3):547-556.
- [32] JANG SH, BYUN JK, JEON WI, *et al.* Nuclear localization and functional characteristics of voltage-gated potassium channel Kv1.3 [J]. *J Biol Chem*, 2015, 290(20):12547-12557.
- [33] BORTNER CD, CIDLOWSKI JA. Ions, the movement of water and the apoptotic volume decrease [J]. *Front Cell Dev Biol*, 2020, 8:611211.
- [34] HOFFMANN EK, PEDERSEN SF. Cell volume homeostatic mechanisms: effectors and signalling pathways [J]. *Acta Physiol (Oxf)*, 2011, 202(3):465-485.
- [35] BLACKISTON DJ, MCLAUGHLIN KA, LEVIN M. Bioelectric controls of cell proliferation: ion channels, membrane voltage and the cell cycle [J]. *Cell Cycle*, 2009, 8(21):3527-3536.
- [36] URREGO D, TOMCZAK AP, ZAHED F, *et al.* Potassium channels in cell cycle and cell proliferation [J]. *Philos Trans R Soc Lond B Biol Sci*, 2014, 369(1638):20130094.
- [37] OUADID-AHIDOUCH H, AHIDOUCH A. K^+ channel expression in human breast cancer cells: involvement in cell cycle regulation and carcinogenesis [J]. *J Membr Biol*, 2008, 221(1):1-6.
- [38] ZHANG M, YIN HJ, WANG WP, *et al.* Over-expressed human TREK-1 inhibits CHO cell proliferation via inhibiting PKA and p38 MAPK pathways and subsequently inducing G1 arrest [J]. *Acta Pharmacol Sin*, 2016, 37(9):1190-1198.
- [39] KRABBENDAM IE, HONRATH B, DILBERGER B, *et al.* SK channel-mediated metabolic escape to glycolysis inhibits ferroptosis and supports stress resistance in *C. elegans* [J]. *Cell Death Dis*, 2020, 11(4):263.
- [40] REINHARDT J, GOLENHOFEN N, PONGS O, *et al.* Migrating transformed MDCK cells are able to structurally polarize a voltage-activated K^+ channel [J]. *Proc Natl Acad Sci U S A*, 1998, 95(9):5378-5382.
- [41] KESSLER W, BUDDE T, GEKLE M, *et al.* Activation of cell migration with fibroblast growth factor-2 requires calcium-sensitive potassium channels [J]. *Pflugers Arch*, 2008, 456(5):813-823.
- [42] GIRAULT A, BROCHIERO E. Evidence of K^+ channel

- function in epithelial cell migration, proliferation, and repair [J]. *Am J Physiol Cell Physiol*, 2014, 306(4): C307-C319.
- [43] ORLOV SN, HAMET P. Intracellular monovalent ions as second messengers [J]. *J Membr Biol*, 2006, 210(3): 161-172.
- [44] SCHWAB A. Function and spatial distribution of ion channels and transporters in cell migration [J]. *Am J Physiol Renal Physiol*, 2001, 280(5): F739-F747.
- [45] KIMURA K, KAWANO S, MORI T, *et al.* Quantitative analysis of the effects of extracellular matrix proteins on membrane dynamics associated with corneal epithelial cell motility [J]. *Invest Ophthalmol Vis Sci*, 2010, 51(9): 4492-4499.
- [46] MUNOZ-PLANILLO R, KUFFA P, MARTINEZ-COLON G, *et al.* K⁺ efflux is the common trigger of NLRP3 inflammasome activation by bacterial toxins and particulate matter [J]. *Immunity*, 2013, 38(6): 1142-1153.
- [47] GAIDT MM, EBERT TS, CHAUHAN D, *et al.* The DNA inflammasome in human myeloid cells is initiated by a STING-cell death program upstream of NLRP3 [J]. *Cell*, 2017, 171(5): 1110-1124.e18.
- [48] SCHULTE-MECKLENBECK A, BITTNER S, EHLING P, *et al.* The two-pore domain K2 P channel TASK2 drives human NK-cell proliferation and cytolytic function [J]. *Eur J Immunol*, 2015, 45(9): 2602-2614.
- [49] FERNÁNDEZ-ORTH J, ROLFES L, GOLA L, *et al.* A role for TASK2 channels in the human immunological synapse [J]. *Eur J Immunol*, 2021, 51(2): 342-353.
- [50] NORTON RS, PENNINGTON MW, WULFF H. Potassium channel blockade by the sea anemone toxin ShK for the treatment of multiple sclerosis and other autoimmune diseases [J]. *Curr Med Chem*, 2004, 11(23): 3041-3052.
- [51] PARDO LA, STUHMER W. The roles of K⁺ channels in cancer [J]. *Nat Rev Cancer*, 2014, 14(1): 39-48.
- [52] LASTRAIOLI E. Focus on triple-negative breast cancer: potassium channel expression and clinical correlates [J]. *Front Pharmacol*, 2020, 11: 725.
- [53] TEISSEYRE A, PALKO-LABUZ A, SRODA-POMIANEK K, *et al.* Voltage-gated potassium channel Kv1.3 as a target in therapy of cancer [J]. *Front Oncol*, 2019, 9: 933.
- [54] STEGEN B, KLUMPP L, MISOVIC M, *et al.* K⁺ channel signaling in irradiated tumor cells [J]. *Eur Biophys J*, 2016, 45(7): 585-598.
- [55] PALME D, MISOVIC M, GANSER K, *et al.* hERG K⁺ channels promote survival of irradiated leukemia cells [J]. *Front Pharmacol*, 2020, 11: 489.
- [56] KAMATHAM S, WATERS CM, SCHWINGSHACKL A, *et al.* TREK-1 protects the heart against ischemia-reperfusion-induced injury and from adverse remodeling after myocardial infarction [J]. *Pflugers Arch*, 2019, 471(10): 1263-1272.
- [57] TINKER A, AZIZ Q, LI Y, *et al.* ATP-sensitive potassium channels and their physiological and pathophysiological roles [J]. *Compr Physiol*, 2018, 8(4): 1463-1511.
- [58] SHIMIZU S, SAITO M, KINOSHITA Y, *et al.* Nicorandil ameliorates ischaemia-reperfusion injury in the rat kidney [J]. *Br J Pharmacol*, 2011, 163(2): 272-282.
- [59] OLDENBURG O, QIN Q, KRIEG T, *et al.* Bradykinin induces mitochondrial ROS generation via NO, cGMP, PKG, and mitoKATP channel opening and leads to cardioprotection [J]. *Am J Physiol Heart Circ Physiol*, 2004, 286(1): H468-H476.
- [60] COSTA AD, GARLID KD. Intramitochondrial signaling: interactions among mitoKATP, PKCepsilon, ROS, and MPT [J]. *Am J Physiol Heart Circ Physiol*, 2008, 295(2): H874-H882.
- [61] CHANDY KG, DECOURSEY TE, FISCHBACH M, *et al.* Altered K⁺ channel expression in abnormal T lymphocytes from mice with the *lpr* gene mutation [J]. *Science*, 1986, 233(4769): 1197-2000.
- [62] OHYA S, KAJIKURI J, ENDO K, *et al.* KCa1.1 K⁺ channel inhibition overcomes resistance to antiandrogens and doxorubicin in a human prostate cancer Lncap spheroid model [J]. *Int J Mol Sci*, 2021, 22(24): 13553.
- [63] ENDO K, KITO H, TANAKA R, *et al.* Possible contribution of inflammation-associated hypoxia to increased K^{2p5.1} K⁺ channel expression in CD4⁺ T cells of the mouse model for inflammatory bowel disease [J]. *Int J Mol Sci*, 2019, 21(1): 38.
- [64] TANNER MR, PENNINGTON MW, LARAGIONE T, *et al.* KCa1.1 channels regulate beta1-integrin function and cell adhesion in rheumatoid arthritis fibroblast-like synoviocytes [J]. *FASEB J*, 2017, 31(8): 3309-3320.
- [65] NAKAKURA S, MATSUI M, SATO A, *et al.* Pathophysiological significance of the two-pore domain K(+) channel K2P5.1 in splenic CD4(+) CD25(-) T cell subset from a chemically-induced murine inflammatory bowel disease model [J]. *Front Physiol*, 2015, 6: 299.
- [66] SCHIRMER L, SRIVASTAVA R, KALLURI SR, *et al.* Differential loss of KIR4.1 immunoreactivity in multiple sclerosis lesions [J]. *Ann Neurol*, 2014, 75(6): 810-828.
- [67] ASHCROFT FM, PULJUNG MC, VEDOVATO N. Neonatal diabetes and the KATP channel: from mutation to therapy [J]. *Trends Endocrinol Metab*, 2017, 28(5):

- 377-387.
- [68] BONFANTI DH, ALCAZAR LP, ARAKAKI PA, *et al.* ATP-dependent potassium channels and type 2 diabetes mellitus[J]. *Clin Biochem*, 2015, 48(7-8): 476-482.
- [69] SEPULVEDA FV, PABLO CID L, TEULON J, *et al.* Molecular aspects of structure, gating, and physiology of pH-sensitive background K2P and Kir K+-transport channels[J]. *Physiol Rev*, 2015, 95(1): 179-217.
- [70] DAVIES LA, HU C, GUAGLIARDO NA, *et al.* TASK channel deletion in mice causes primary hyperaldosteronism [J]. *Proc Natl Acad Sci U S A*, 2008, 105(6): 2203-2208.
- [71] ZHANG X, WAN JQ, TONG XP. Potassium channel dysfunction in neurons and astrocytes in Huntington's disease[J]. *CNS Neurosci Ther*, 2018, 24(4): 311-318.
- [72] ZHANG L, ZHENG Y, XIE J, *et al.* Potassium channels and their emerging role in parkinson's disease[J]. *Brain Res Bull*, 2020, 160: 1-7.
- [73] VILLA C, SUPHESIZ H, COMBI R, *et al.* Potassium channels in the neuronal homeostasis and neurodegenerative pathways underlying Alzheimer's disease: An update [J]. *Mech Ageing Dev*, 2020, 185: 111197.
- [74] TONG X, AO Y, FAAS GC, *et al.* Astrocyte Kir4.1 ion channel deficits contribute to neuronal dysfunction in Huntington's disease model mice[J]. *Nat Neurosci*, 2014, 17(5): 694-703.
- [75] GUPTA S, SHARMA B. Protective effects of phosphodiesterase-1 (PDE1) and ATP sensitive potassium (KATP) channel modulators against 3-nitropropionic acid induced behavioral and biochemical toxicities in experimental Huntingtons disease [J]. *Eur J Pharmacol*, 2014, 732: 111-122.
- [76] CANG C, ARANDA K, SEO YJ, *et al.* TMEM175 is an organelle K(+) channel regulating lysosomal function [J]. *Cell*, 2015, 162(5): 1101-1112.
- [77] WIE J, LIU Z, SONG H, *et al.* A growth-factor-activated lysosomal K⁺ channel regulates Parkinson's pathology [J]. *Nature*, 2021, 591(7850): 431-437.

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- [24] 上海市卫生健康委员会. 4月28日上海通报新冠肺炎防控情况[EB/OL]. (2022-04-28) [2022-04-29]. <http://wsjkw.sh.gov.cn/xwfbh/20220428/bcf13c1b93a4457f89b2b541fc1dd228.html>.
- [25] KONISHI S, KITAGAWA G. Information criteria and statistical modeling[M]. New York: Springer-Verlag, 2008.
- [26] GNEITING T, BALABDAOUI F, RAFTERY AE. Probabilistic forecasts, calibration and sharpness [J]. *J R Stat Soc B*, 2007, 69(2): 243-268.
- [27] CZADO C, GNEITING T, HELD L. Predictive model assessment for count data [J]. *Biometrics*, 2009, 65(4): 1254-1261.
- [28] FUNK S, CAMACHO A, KUCHARSKI AJ, *et al.* Assessing the performance of real-time epidemic forecasts: a case study of Ebola in the Western Area region of Sierra Leone, 2014-15 [J]. *PLoS Comput Biol*, 2019, 15(2): e1006785.
- [29] CARPENTER B, GELMAN A, HOFFMAN M D, *et al.* Stan: a probabilistic programming language [J]. *J Stat Softw*, 2017, 76(1): 1-32.
- [30] RSTUDIO. Shiny: web application framework for R. package version 1.7R.1 [EB/OL]. (2021-10-01) [2022-02-19]. <https://shiny.rstudio.com/>.
- [31] 刘芷希, 朱文龙, 王伟炳. 基于时变再生数的上海市新型冠状病毒奥密克戎疫情趋势评估[J]. *上海预防医学*, 2022, 34(6): 541-544.
- [32] 刘可伋, 江渝, 严阅, 等. 局部新冠肺炎时滞模型及再生数的计算[J]. *控制理论与应用*, 2020, 37(3): 453-460.
- [33] 上海市卫生健康委员会. 有序组织、上门接种, 这些区加快推进老年人新冠疫苗接种[EB/OL]. (2022-04-27) [2022-05-11]. <http://wsjkw.sh.gov.cn/xwfb/20220428/55ae7b4d74584b478e6a1b51f2d4cc99.html>.
- [34] 黄伟, 王雅洁, 吴洪宇, 等. 基于大数据的新冠疫情研判预测系统设计与实现[J]. *数字技术与应用*, 2021, 39(8): 148-151.
- [35] 邢策梅, 陶金梅, 范娟娟, 等. 基于微服务架构的江苏省新冠疫情大数据平台的设计与实现[J]. *现代测绘*, 2020, 43(3): 34-38.

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